

WHAT IS CLAIMED IS:

1. A method for introducing an expression cassette into a target cell of a vascularized multi-cellular organism in a manner such that the encoded protein of said expression cassette is persistently expressed in said target cell at a high level, said method comprising:  
5 systemically administering to said vascularized multi-cellular organism a minimal plasmid vector comprising said expression cassette, wherein said minimal plasmid vector provides for persistent and high level expression in a manner that is substantially expression cassette sequence and direction independent;  
10 to persistently express said expression cassette encoded protein at a high level in said target cell.
2. The method according to Claim 1, wherein said administering is intravenous.
- 15 3. The method according to Claim 1, wherein said vascularized multi-cellular organism is a mammal.
4. The method according to Claim 1, wherein said minimal plasmid vector further comprises an antibiotic resistance gene.  
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5. The method according to Claim 1, wherein said minimal plasmid vector further comprises a multiple cloning site.
6. The method according to Claim 1, wherein said minimal plasmid vector further  
25 comprises a plasmid origin of replication.
7. The method according to Claim 1, wherein said target cell is hepatic cell.
8. A method of expressing a protein in a target cell of a mammal, said method  
30 comprising:

intravenously administering to said mammal an aqueous formulation of a minimal plasmid vector comprising an expression cassette encoding said protein, wherein said minimal plasmid vector provides for persistent and high level expression in a manner that is substantially expression cassette sequence and direction independent;

5           whereby said expression cassette encoded protein is expressed in said target cell.

9.       The method according to Claim 8, wherein said minimal plasmid vector further comprises an antibiotic resistance gene.

10       10.     The method according to Claim 8, wherein said minimal plasmid vector further comprises a multiple cloning site.

11.     The method according to Claim 8, wherein said minimal plasmid vector further comprises a plasmid origin of replication.

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12.     The method according to Claim 8, wherein said target cell is a hepatic cell.

13.     A method of persistently expressing a protein at a high level in a hepatic target cell of a mammal, said method comprising:

20           intravenously administering to said mammal an aqueous formulation of a minimal plasmid vector comprising an expression cassette encoding said protein, wherein said minimal plasmid vector provides for persistent and high level expression in a manner that is substantially expression cassette sequence and direction independent;

25           whereby said expression cassette encoded protein is persistently expressed at a high level in said hepatic target cell.

14.     The method according to Claim 13, wherein said minimal plasmid vector further comprises an antibiotic resistance gene.

30       15.     The method according to Claim 13, wherein said minimal plasmid vector further comprises a multiple cloning site.

16. The method according to Claim 13, wherein said minimal plasmid vector further comprises a plasmid original of replication.
- 5 17. A minimal plasmid vector that provides for persistent and high level expression of an expression cassette present therein in a manner that is substantially expression cassette sequence and direction independent.
18. The minimal plasmid vector according to Claim 17, wherein said vector further  
10 comprises a multiple cloning site.
19. The minimal plasmid vector according to Claim 18, wherein said vector further comprises a plasmid origin of replication.
- 15 20. The minimal plasmid vector according to Claim 17, wherein said vector further comprises an expression cassette.
21. A pharmaceutical composition comprising as an active agent a minimal plasmid vector that provides for persistent and high level expression of an expression cassette present  
20 therein in a manner that is substantially expression cassette sequence and direction independent together with a pharmaceutically acceptable carrier, diluent and/or adjuvant.
22. The composition of Claim 21 for expression of a heterologous nucleic acid in a vascularized multi-cellular organism.  
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23. The composition of Claim 21, which is administered systemically or locally.
24. The composition of Claim 21, for gene therapy.
- 30 25. The composition of Claim 21, for nucleic acid vaccination.

26. The use of a minimal plasmid vector that provides for persistent and high level expression of an expression cassette present therein in a manner that is substantially expression cassette sequence and direction independent for the manufacture of an agent for heterologous gene expression in a vascularized multi-cellular organism.

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